

**SEASONAL VARIATIONS IN THE ABUNDANCE OF PHYTOPLANKTON  
IN THE SHALLOW WATERS OF CUA BE RIVER ESTUARY,  
NHA TRANG BAY, CENTRAL VIETNAM**

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**ABSTRACT** Thirty-seven species of dinoflagellates and 60 species of diatoms were the main components of the phytoplankton community in the shallow waters of Cua Be river estuary. The key species in the study area in terms of both biomass and frequency of occurrence were *Gonyaulax* spp., *Protoperdinium* spp., and *Peridinium quinquecorne* (dinoflagellates), and centric diatom *Coscinodiscus* spp., *Skeletonema costatum* and *Rhizosolenia* spp. The species composition changed markedly between the dry and rainy seasons. Temperature and salinity were the two main factors affecting the seasonal shifts of species. Peaks of diatoms and dinoflagellates occurred together. Dinoflagellates were abundant throughout the year with highest concentrations in the middle of the dry season and at the beginning of the rainy season. Concentrations of nutrients such as nitrate-nitrogens and ortho-phosphate-phosphorus were strongly related with salinity, but the relations between dinoflagellates and diatoms and nutrients were not clear.

**BIẾN ĐỘNG THEO MÙA CỦA THỰC VẬT PHUẾ DU TRONG THUY VỐC  
NƯỚC NÔNG VÙNG CỎA SÔNG CỎA BỂ VỒNH NHA TRANG,  
MIỀN TRUNG VIỆT NAM**

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**TÓM TẮT** 37 loài Tảo Hai Roi và 60 loài Tảo Silíc là thành phần chủ yếu của quần xã Thực Vật Phú Du trong thuy vốc nước nông ven bờ vùng cửa sông Cửa Bể. Các loài chủ yếu quyết định sinh khối cũng như tần số xuất hiện là các loài Tảo Giáp Hai Roi *Gonyaulax* spp., *Protoperdinium* spp. và Tảo Silíc Trung tâm *Coscinodiscus* spp., *Skeletonema costatum* và *Rhizosolenia* spp. Thành phần loài thay đổi rõ rệt trong hai mùa khô và mùa mưa. Nhiệt độ và độ mặn là 2 yếu tố chính ảnh hưởng nên những thay đổi về mùa vụ. Nồng độ của Tảo Silíc cũng là nhân tố chính của Tảo Hai Roi. Tảo Hai Roi phong phú quanh năm với mật độ cao nhất vào giữa mùa khô và đầu mùa mưa. Hàm lượng muối dinh dưỡng như  $SiO_3-Si$ ,  $NO_3-N$  và  $PO_4-P$  có liên quan chặt chẽ với độ mặn của nước, những mối quan hệ giữa các thành phần sinh học (Tảo Hai Roi và Tảo Silíc) và các muối dinh dưỡng không rõ ràng.

## I. INTRODUCTION

The study of planktonic organisms in Vietnamese waters began after the establishment of the Institute of Oceanography in 1923, in Nha Trang city (Nguyen-Van et al. 1995). The phytoplankton in Nha Trang bay has therefore received considerable scientific attention (Rose 1926, 1955, Hoang 1962, 1963, Shirota 1963, 1966). A major contribution was made by Prof. Hoang Quoc Truong, who described and illustrated 154 species of diatoms and 92 species of dinoflagellates in Nha Trang bay (Hoang 1962, 1963). Since then many phytoplankton surveys in the bay have been conducted, with most listing species composition over a short period either in the rainy or dry seasons.

Since the 1990s, there has been rapid expansion of coastal development in the Cua Be river estuary, including aquaculture, tourist services, agriculture and the increase of population in this area, with sewage inputs through lack of waste treatment services. These and other activities were thought to have played an important role in the structure and seasonal changes of the phytoplankton community in the Nha Trang bay, although no prior studies had demonstrated this. For example, blooms of cyanobacteria *Trichodesmium erythraeum* and *T. thiebautii*, dinoflagellates such as *Gonyaulax* spp., *Peridinium quinquerorne* were sometimes observed in the bay from 1998 to 2002 (pers. observ.).

Most previous phytoplankton surveys in Nha Trang bay were not conducted through a sufficient time period to allow analysis of ecological aspects of seasonal variation, including changes / succession in species

composition, or the relationship between phytoplankton and environmental factors (e.g. nutrients, salinity, and temperature). The present study addressed these aspects by monitoring the seasonal variation in abundance of phytoplankton species, especially of dinoflagellates, at one anchored station in the estuary of the Cua Be river.

## II. MATERIALS AND METHODS

### 1. Description of sampling locality

The sampling site was located in the Cua Be river estuary (Fig. 1), strongly affected by fresh water discharge during the rainy season and seawater in the dry season /or in the spring tidal period. Marine aquaculture and other human activities were rapidly increasing along the coast, contributing to change of the biological components in the area. Most shrimp aquaculture activities occur in the dry season, notably from January to September each year.

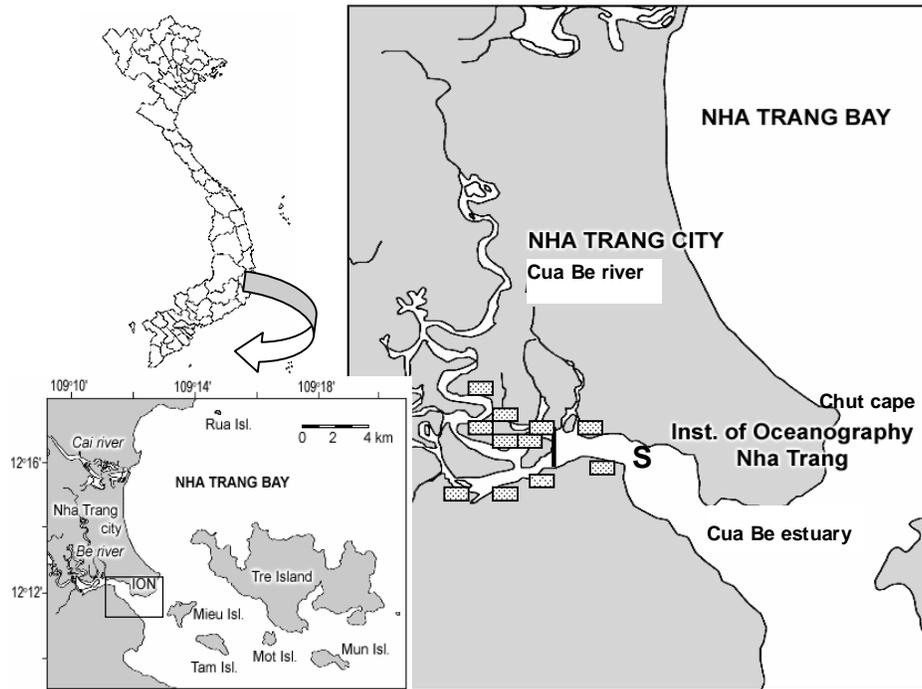
On average, the area receives more than 2000 mm of rain annually, with the highest total per month (>500 mm) in October-November (Northeast Monsoon), decreasing in December and with less than 200 mm of rainfall per month during the remaining months (Southwest Monsoon, Fig. 2). Salinity in the estuary is strongly affected by rainfall and river run-off. In Nha Trang bay generally, salinity ranges from 33-35 psu in the dry season and decreases to 25-28 psu in the rainy season. Mean water temperature ranges between 28-29°C in the dry season (max. 31.4°C), and 21-22°C in the rainy season (max. 23-24°C) (Dawydoff 1931, 1933, Serene 1935, 1949).

### 2. The sampling programme

Samples were collected weekly during the period from March 10<sup>th</sup> 2000 to Feb. 23<sup>rd</sup> 2001 between 8 and 9 a.m.

At the sampling station (Fig. 1), vertical hauls were taken with a 20 µm mesh plankton net, and water samples

of 1 litre were collected at the surface with a bottle or a water sampler. All samples were fixed in neutral Lugol's solution, stored cold and dark, and transported to the nearby laboratory at Institute of Oceanography.



**Fig. 1:** Map showing the position of the Cua Be estuary and sampling station (S), note the marine aquaculture ponds along the river bank (dotted rectangles)

Temperature and salinity were measured using a hand CTD and a salinity refractometer with a precision of 1 psu. Rainfall data for the study period were provided by the Meteorological Stations of Nha Trang City. Nutrients (e.g. nitrate - nitrogens, ortho - phosphate - phosphorus, and silicate - silica) and pigments (e.g. chlorophyll-a and pheophytine) from the water samples were analyzed by Dept. of Hydro - geochemistry of Institute of Oceanography, Nha Trang, according to APHA (1992).

### 3. Taxonomic analyses

Dinoflagellate plate patterns were stained with Calcofluor White M2R (Fritz & Triemer 1985), sometimes in combination with the use of hypochlorite. The samples were examined on an epifluorescence (violet excitation c. 430 nm, blue emission c. 490 nm) Leica DMLB microscope with phase contrast and differential interference contrast optics. For identification purposes, the plate patterns in the two orders Peridiniales and Gonyaulacales were usually described using the Kofoidian system

(Kofoid 1911). Kodak Technical Pan film was used for photography.

The identification of phytoplankton followed Abell (1936), Schiller (1933, 1937), Lebour (1925), Kofoid (1911), Kofoid & Skogsberg (1928), Taylor (1976), and Steindinger (1997) for dinoflagellate organisms; and Allen & Cup (1935), Cup (1943), Hasle (1965), Desikachary (1986 - 1989), Hargraves (1979), Hernandez - Becerril (1996), Tomas (1997) and Jensen & Moestrup (1998) for diatom frustules. Most dinoflagellates and diatoms were identified to species level, except for Pennate diatoms (*Pseudo-nitzschia*, *Nitzschia*, *Navicula*, *Diploneis*, etc.), which were identified to genus or higher taxonomic group.

#### **4. Cell counts and estimation of biomass**

The water samples were concentrated in the laboratory and cell counts were carried out on an inverted microscope using a 1 ml Sedgewick-Rafter cell. Thecate dinoflagellates were counted by epifluorescence microscopy following Andersen & Kristensen (1995). Cell dimensions of species were measured following Edler (1979)

#### **5. Data analyses**

Cell volumes, biomass, and carbon contents per cell were calculated according to the geometric formulas and conversion factors given in Edler (1979).

Phytoplankton abundance is expressed as the logarithm of the number of cells per liter plus 1 [ $\log(x+1)$ ].

All raw data were stored and analyzed in the database, ALGAESYS (Bio/consult Ltd.), using the statistical package PRIMER (Plymouth Marine Laboratory, 1994). Bray-Curtis simi-

larity index was used for analysis of structure of species composition between months/seasons. Principal Components Analysis was used to explore the relationships among species /or species group and environmental variables (salinity, temperature, nutrients).

### **III. RESULTS**

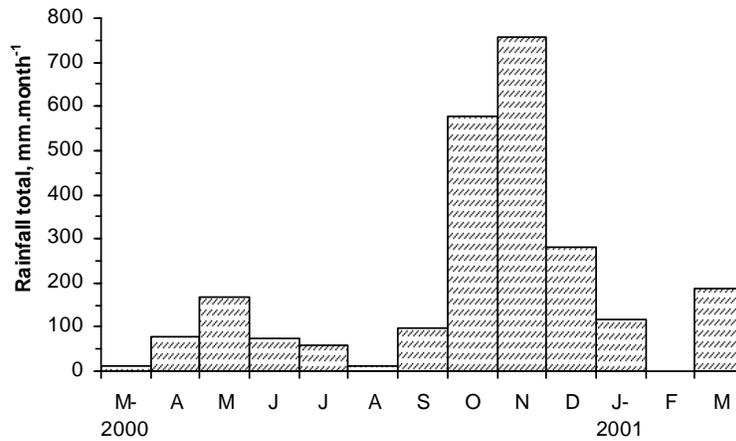
#### **1. Salinity and temperature**

Cua Be river estuary experiences two major bi-annual periods of different thermohaline characteristics (Fig. 3), corresponding to the dry and rainy seasons respectively. During the dry season (March to the end of August), Cua Be river estuary was characterized by relatively high salinities, above 30 psu. The rainy season, starting in September 2000, was characterized by lower salinity levels, reaching a seasonal low (< 10 psu) in November, before gradually increasing again. The weekly variations in salinity (saw-like pattern of the curve) was likely related with the phasing of the tidal cycle at the time of each sampling and its interactions with the seasonal variations of Cua Be river flow regime. The strong influence of inland freshwater run-off during the period of end of August until February of the following year was clearly apparent in the samples (c.f. Figs 2 and 3).

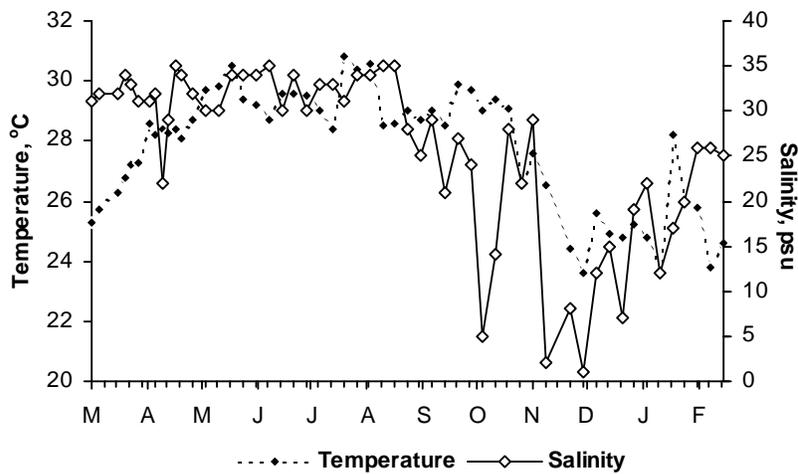
During this present study, water temperatures ranged from 23.8°C to 30.8°C with annual average of 27.7 ± 1.9°C. Temperatures in December and January-February were the lowest in the year (24.7 ± 0.80°C, and 25.3 ± 1.4 °C respectively), whereas the highest temperature occurred in May (29.7± 0.7°C). The average temperature in the period of the rainy season Northeast

Monsoon ( $26.2 \pm 1.8^\circ\text{C}$ ) was lower than that in the period of the dry season

Southwest Monsoon, ( $29.4 \pm 0.7^\circ\text{C}$ ).



**Fig. 2:** Monthly rainfall total of Nha Trang area from March 2000 to March 2001



**Fig. 3:** Variation in temperature and salinity, Cua Be river estuary, Vietnam 2000-2001

Observations on the variation in temperature and salinity and based upon the monthly rainfall chart showed that there were two main seasons in study area, the dry season with low rainfall and high salinity from January to September and the rainy season with low salinity during the remaining months. However, in the dry season, which could be divided into two main periods: - a cold period with low

salinity from mid-December to February, average temperature  $26.5 \pm 2.1^\circ\text{C}$  and average salinity  $17.8 \pm 8.8$  psu, this period could be divided in two sub-periods, one sub-period with high temperature  $27.8 \pm 1.8^\circ\text{C}$  (Oct. - Nov.) and an other one with low temperature  $25.2 \pm 1.2^\circ\text{C}$  (mid-Dec. - Feb.); and - a warm period with high average salinity,  $31.5 \pm 1.8$  psu, starting from March to early December.

## 2. Nutrients

Nitrate-nitrogen ( $\text{NO}_3\text{-N}$ ):

Mean concentrations of nitrate-nitrogen were  $149.0 \pm 69.3 \mu\text{g.l}^{-1}$  (N=29) during the first half of the sampling period from March to August and  $204.0 \pm 47.0 \mu\text{g.l}^{-1}$  (N=25) during the second half (from Sep. onwards). Concentration increased markedly in May (from ca.  $100 \mu\text{g.l}^{-1}$  to  $> 200 \mu\text{g.l}^{-1}$ ), coincident with onset of rainfall (Fig. 2), and remained high ( $> 150 \mu\text{g.l}^{-1}$ ) till October before a sharp decline (ca.  $120 \mu\text{g.l}^{-1}$ ), and more gradual increase to high levels ( $> 200 \mu\text{g.l}^{-1}$ ). The October slump in nitrate-nitrogen immediately preceded the peak in phytoplankton biomass, suggesting rapid uptake of the nutrients (see later).

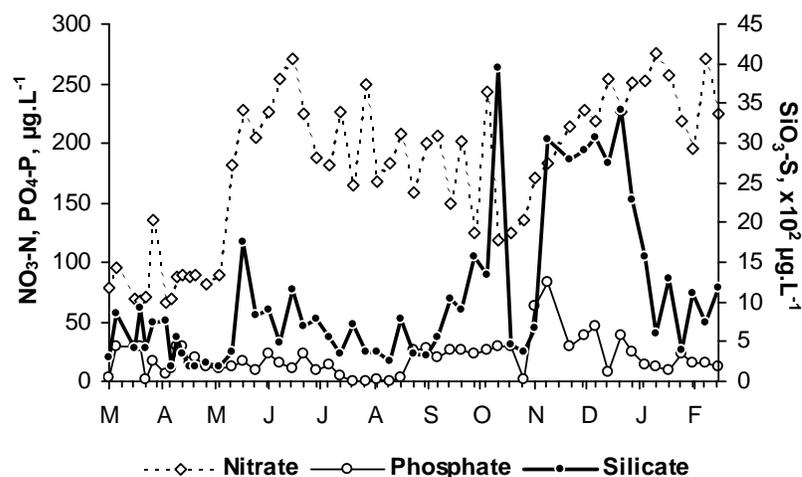
Relative peaks of nitrate-nitrogen concentrations appear to coincide with periods of lower salinity, indicating their fluvial origin, being lowest during March and April of 2000 (dry season) with a strong increase during the summer months (May – August), with onset of rainfall in May (Fig. 2), remaining at high levels ( $> 150 \mu\text{g.l}^{-1}$ )

through most of the rainy season and winter. The strong increase in nitrate-nitrogen concentrations in May 2000 is likely to be related to the combined impact of the May rainfall and shrimp pond culture effluents pouring into the Cua Be river.

Ortho-phosphate-phosphorus ( $\text{PO}_4\text{-P}$ ):

Mean concentrations of ortho-phosphate-phosphorus during the first half of the sampling period (Mar. – Aug. 2000) were  $13.5 \pm 9.9 \mu\text{g.l}^{-1}$ , increasing to  $26.7 \pm 17.4 \mu\text{g.l}^{-1}$  during the second half. Ortho - phosphate - phosphorus showed less variation in concentration than nitrate-nitrogen, but exhibited a broadly similar trend in temporal variation in concentration. Peaks in concentration also coincided with periods of low salinity, with a strong peak (reaching  $80 \mu\text{g.l}^{-1}$ ) associated with one of the lowest salinity periods (November 2000), and conversely very low ortho-phosphate-phosphorus concentrations coincided with high salinities (e.g. in Jul. and Aug. 2000).

Silicate-silica ( $\text{SiO}_3\text{-Si}$ ):



**Fig. 4:** Variation in nutrient concentration (nitrate-nitrogen, ortho-phosphate-phosphorus, and silicate-silica), Cua Be river estuary, Vietnam 2000-2001

Mean values of silicate-silica during the first half of the sampling period were  $574.3 \pm 352 \mu\text{g.l}^{-1}$ , increasing to  $1546.3 \pm 1125.3 \mu\text{g.l}^{-1}$  during the second half. Silicate - silica concentration exhibited similar trends to orthophosphate-phosphorus and to a lesser extent with nitrate-nitrogen, with high concentrations during the rainy season when salinity values were lower.

All three nutrients exhibited rapid precipitous decline in concentration in October 2000, immediately preceding highest phytoplankton biomass, suggesting rapid uptake (see later).

### 3. Species composition

100 species of phytoplankton from five algal groups: cyanobacteria, dinoflagellates, chrysophytes, diatoms and chlorophytes were present at the sampling site in the Cua Be river estuary during 2000-2001. Predominant among this phytoplankton flora were

38 species of dinoflagellates and 63 species of diatoms (Appendix tab. 1), the common species being the dinoflagellates, *Gonyaulax verior*, *G. spinifera*, *G. polyedra*, and *Peridinium quinquecorne*, and diatoms *Chaetoceros* spp., *Coscinodiscus* spp., *Pleurosigma*, and *Pseudo-nitzschia* spp.

There were strong seasonal trends in species composition with highest diversity (richness) in the dry season (Fig. 5). Some species characteristic of freshwater habitats, such as *Oscillatoria* sp. (Cyanobacteria) and *Scenedesmus* sp. (Chlorophytes), were present in the rainy season, disappearing from the sampling site in the dry season. Conversely a suite of species was present in the dry season, some of which are listed above (also see Appendix tab. 1). Some species were present throughout the year, but with clear seasonal trends in abundance (e.g. Diatoms *Coscinodiscus* spp. and Dinoflagellates *Gonyaulax* spp.).

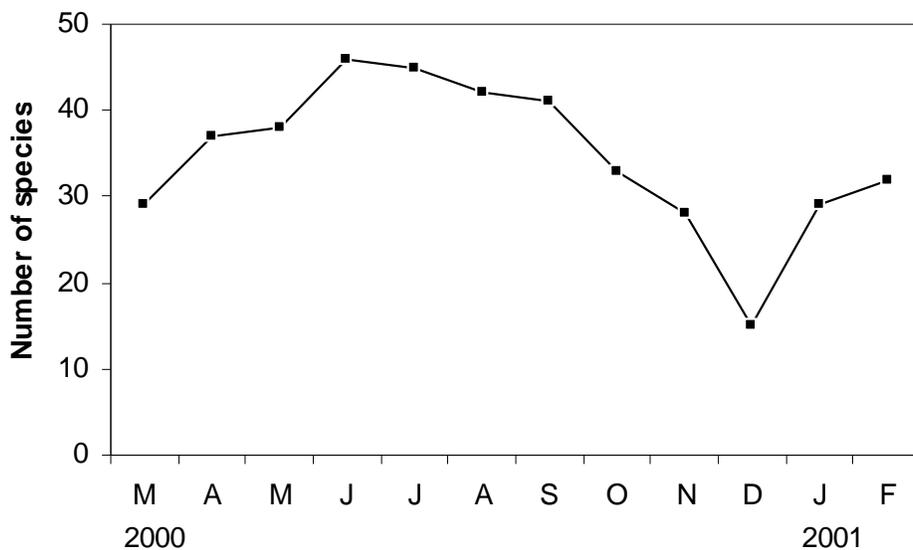


Fig. 5: Species diversity (richness) of phytoplankton, Cua Be river estuary 2000-01

There was strong similarity in species composition and fidelity to the different seasons, with two major species groups, each with two sub-groups (Fig. 6):

- **Species groups 1** in the rainy season (RS)-low salinity and low temperature during Oct.-Dec. & Jan.-Feb., in which:

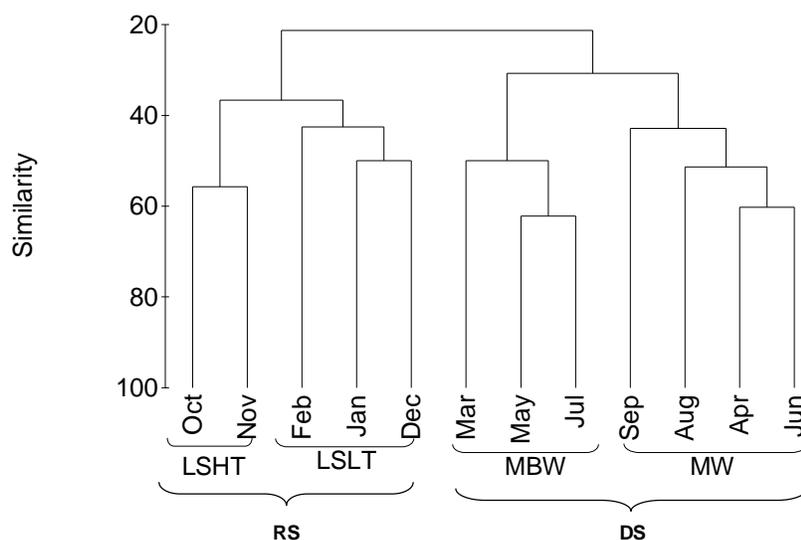
- species sub-group I of very low salinity (fresh water) and high temperature (LSHT) in October-November 2000.

- species sub-group II of low salinity (brackish water) and low temperature (LSLT) in Dec. 2000 – Feb. 2001.

- **Species group 2** in the dry season (DS)-high salinity and high temperature during Mar.-Sep. 2000, in which:

- species sub-group III of marine & brackish water (MBW) in Mar., May, Jul. 2000.

- species sub-group IV of marine water (MW) in Apr., Jun., Aug., and Sep. 2000.



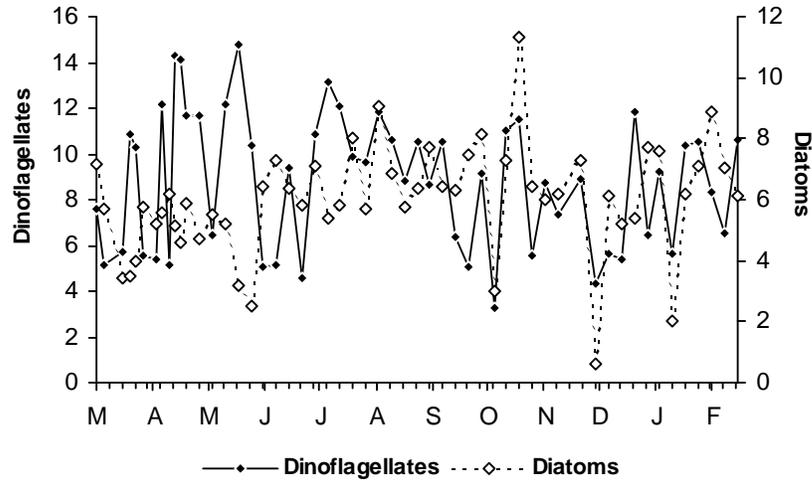
**Fig. 6:** Similarity in phytoplankton community structures (species composition) in Cua Be River estuary 2000-01 expressed by Bray-Curtis similarity index.

#### 4. Cell concentration

Phytoplankton total: Average cell concentration was  $218 \times 10^3 \text{ cells.l}^{-1}$ . Although there was no different in average cell concentration between the dry season and the rainy season overall, ( $\text{ca.}210 \times 10^3 \text{ cells.l}^{-1}$ ) the average value was the highest during the high temperature period of the dry season from Mar. to Aug. ( $\text{ca.}260 \times 10^3 \text{ cells.l}^{-1}$ ) and was very low ( $\text{ca.}50 \times 10^3 \text{ cells.l}^{-1}$ ) during the low temperature

period from Dec. to Feb. 2001. Average cell concentration of dinoflagellates was twice that of diatoms (Fig. 7).

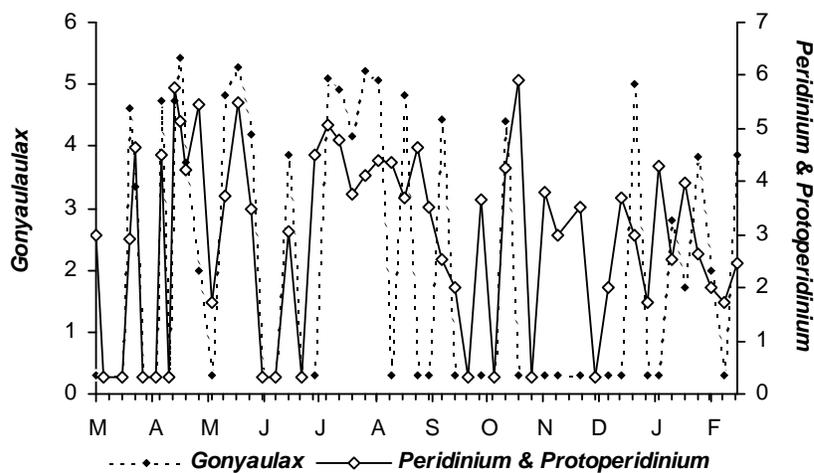
Dinoflagellates: As noted above, some species of dinoflagellates were present all year round, especially during the summer months (Apr.-Aug.). Among them two major groups: *Gonyaulax* spp. and species in the Order Peridinales dominated cell concentration and biomass during the year.



**Fig. 7:** Monthly abundance of dinoflagellates and diatoms. Note the concentration (cells.l<sup>-1</sup>) transformed into log (x+1)

- *Gonyaulax* species were particularly abundant in periods of relatively high salinity and showed lowest densities during the rainy season (Oct.-Dec.). *Gonyaulax* spp. may have potential as an indicator of seawater intrusion in the estuary, as the saw-like pattern of their abundance appeared to be related to the phasing of the tidal cycle at the time of sampling (Fig. 8).

- *Protoperidinium* and *Peridinium* species were also present throughout the year, even during the rainy season, but exhibiting particular seasonal peaks in abundance during the summer months (Fig. 8). Their highest cell concentrations were in Jul. and Oct. (> 1000 x 10<sup>3</sup> cells.l<sup>-1</sup>) co-dominant with *Gonyaulax* spp. and species of *Peridinium* and *Protoperidinium*.



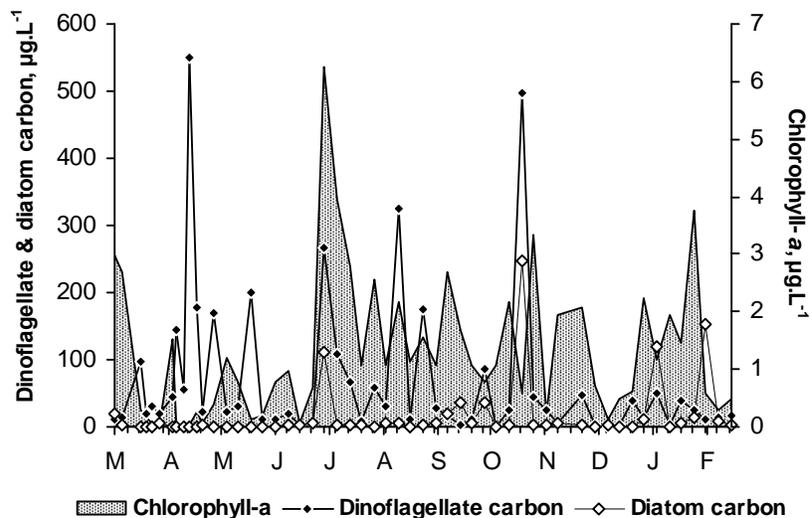
**Fig. 8:** Monthly abundance of two major dinoflagellate groups (*Gonyaulax* spp. and *Peridinales*). Note the concentration (cells.l<sup>-1</sup>) transformed into log (x+1)

Diatoms: The highest concentration (ca.  $1000 \times 10^3 \text{ cell.l}^{-1}$ ) occurred in the dry season (28 Jul. 2000) and the rainy season (27 Oct. 2000), dominated by centric diatoms of large size, such as *Coscinodiscus* spp. and *Rhizosolenia* spp. (Fig. 7).

There were only relatively small seasonal variations in overall concentrations of carbon and chlorophyll-a between the dry and rainy seasons (Tab. 6). By contrast dinoflagellates and diatoms showed more marked seasonal fluctuations, with very high cell concentrations in the dry season and in the rainy season, respectively. Concentrations of phytoplankton carbon and chlorophyll-a are not only dependent on cell concentration, but also on the cell volume. Small size dinoflagellates, such as *Gonyaulax* spp. and *Proto-peridinium* spp., played a decisive role in the dry season, while species of *Coscinodiscus* were responsible for the high biomass in the rainy season.

## 5. Biomass

Average total biomass of phytoplankton was ca.  $85 \mu\text{gC.l}^{-1}$ , and showed a similar seasonal trend with cell concentration (cf. Figs 9 and 7). Dinoflagellate carbon generally was high in the dry season with the highest value  $550 \mu\text{gC.l}^{-1}$  at 21 Apr. 2000. A second peak in biomass (ca.  $500 \mu\text{gC.l}^{-1}$ ) occurred in the rainy season at 27 Oct. 2000. Both these cases were due to the very strong development of *Gonyaulax* spp., *Proto-peridinium* spp. and *Peridinium quinquecorne*. Diatom carbon concentration was high in the rainy season, dominated by centric diatoms such as *Coscinodiscus* spp., *Skeletonema costatum* and *Rhizosolenia* spp. Additionally many small pennate diatoms were present during the rainy season, but they were not responsible for major increases of carbon concentration. Other groups of phytoplankton were also present during the rainy season in the estuary (e.g. cyanobacteria *Oscillatoria* sp. and chlorophytes *Scenedesmus* sp.) but had no influence on phytoplankton concentration and biomass in the samples.



**Fig. 9:** Monthly variation in biomass (carbon concentration) of dinoflagellates and diatoms and chlorophyll-a concentration

## 6. Phytoplankton pigments

Chlorophyll a (Chl-a) concentration was highest during the summer months, peaking in Jul. at  $5.84 \mu\text{g.l}^{-1}$  (Fig. 9). This peak was attributable to high concentrations of dinoflagellates in the dry season. Although not illustrated, pheophytin pigments showed similar trends in concentration as Chl-a, but with higher maximum values ( $6.31 \mu\text{g.l}^{-1}$ ) than Chl-a during the summer months, especially in Sep.

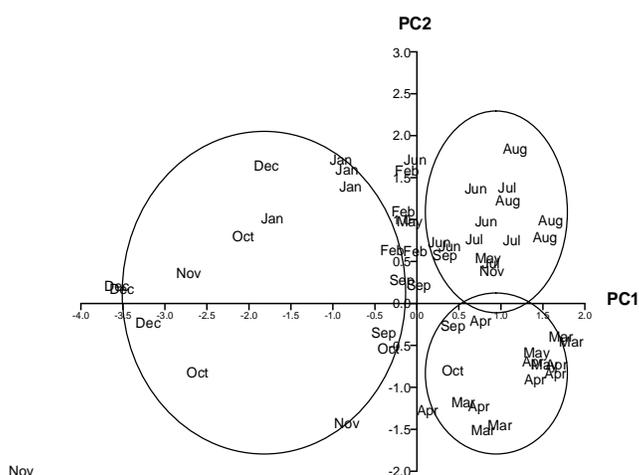
## 7. Relationships between nutrients and salinity

There was a strong seasonal relationship between concentrations of

salinity and nutrients (Fig. 10). The first axis (PC1), contributing 58% of the total variance, relates primarily to salinity variations (Tab. 2), with strong separation between the dry season and the rainy season, the latter characterized by high river flow causing lower salinity. The analysis also demonstrated a negative correlation between salinity and nutrients, particularly silicate (Tab. 2). The second axis (PC2), which together with PC1 explains 83.2% of the total variance, is mostly related to variations in nitrate-nitrogen concentrations, which also show strong seasonal variations (Tab. 2, Fig. 10).

**Table 2:** PCA on the environmental variables showing eigenvalues, % variation and eigenvector coefficient in the linear combinations of variable making up PC axes

PC	Eigen-values	% variation	Cum. % variation	NO3-N	PO4-P	SiO3-Si	Salinity
1	2.3	58.0	58.0	-0.30	-0.45	-0.59	0.60
2	1.0	25.2	83.2	0.83	-0.56	0.01	0.00
3	0.4	10.8	94.0	-0.47	-0.68	0.48	-0.28
4	0.2	6.0	100.0	-0.07	-0.11	-0.65	-0.75



**Fig. 10:** PCA of the abundance of nutrients, and salinity. Plot of two first principal components

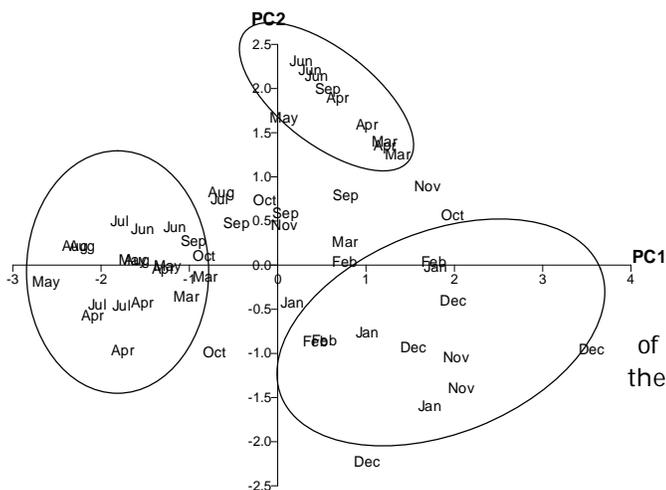
### 8. Relationships between cell concentration of *Gonyaulax* spp., *Peridinium*, *Proto-peridinium* and thermohaline factors

There was also a seasonal relationship between concentrations of dinoflagellates and thermohaline conditions in the estuary (Fig. 11). The first axis (PC1), contributing 51.5% of the total variance, represented seasonal variations in dinoflagellate abundance (Tab. 3). The second axis (PC2), which together with PC1 explained approximately 77% of the total variance, accounted for variations in temperature and salinity during the study period (Tab. 3, Fig. 11).

A correlation matrix between environmental variables (salinity and temperature), nutrients and biological factors was derived from the principal component analysis R-mod (Tab. 4). It showed negative seasonal correlations between salinity and temperature and nutrients. Ortho-phosphate-phosphorus, Nitrate-nitrogen and silicate-silica concentrations gradually increased in the rainy season, coincident with declining salinity and increasing temperature. (Also see Figs 4 & 10). However, the correlative coefficients between the environmental variables and biological factors were relatively weak and unclear.

**Table 3:** PCA on the thermo-haline and dinoflagellates variables showing eigenvalues, % variation and eigenvector coefficient in the linear combinations of variable making up PC axes

PC	Eigen-values	% variation	Cum. % variation	Gonyaulax	Peridinia-les	Tempera-ture	Salinity
1	2.06	51.5	51.5	-0.51	-0.49	-0.51	-0.50
2	1.02	25.4	76.9	-0.46	-0.54	0.49	0.51
3	0.49	12.3	89.2	-0.53	0.48	0.53	-0.46
4	0.43	10.8	100.0	-0.50	-0.49	0.48	-0.53



**Fig. 11:** PCA of the abundance of *Gonyaulax* spp., *Peridiniales*, and thermohaline factors. Plot of two first principal components

**Table 4:** Matrix of correlation coefficient between environmental and biological factors

	Time	Gon	PropPer	DinTotal	DiaTotal	NO <sub>3</sub> -N	PO <sub>4</sub> -P	SiO <sub>3</sub> -Si	Chl-a	Temp	Sal.
Time	1	-	-	-	-	-	-	-	-	-	-
Gon	-0.24	1	-	-	-	-	-	-	-	-	-
PropPer	-0.12	<u>0.49</u>	1	-	-	-	-	-	-	-	-
DinTotal	-0.23	<b>0.69</b>	<b>0.86</b>	1	-	-	-	-	-	-	-
DiaTotal	0.25	-0.14	0.12	0.07	1	-	-	-	-	-	-
NO <sub>3</sub> -N	<b>0.67</b>	-0.04	-0.16	-0.23	0.1	1	-	-	-	-	-
PO <sub>4</sub> -P	0.22	<u>-0.30</u>	-0.17	-0.17	-0.12	-0.01	1	-	-	-	-
SiO <sub>3</sub> -Si	<u>0.49</u>	-0.11	-0.13	-0.21	-0.05	<u>0.33</u>	<u>0.48</u>	1	-	-	-
Chl-a	0.12	-0.03	0.01	0.03	0.16	0.11	-0.19	-0.1	1	-	-
Temp	<u>-0.48</u>	0.27	<u>0.31</u>	<u>0.34</u>	0.27	-0.2	-0.25	<u>-0.39</u>	0.05	1	-
Sal	<b>-0.66</b>	<u>0.30</u>	0.24	<u>0.37</u>	0.13	<u>-0.36</u>	-0.53	<b>-0.76</b>	-0.01	0.54	1

- Time = sampling time/samples; Gon = Gonyaulax spp., PropPer = Peridinales (Protoperidinium & Peridinium), DinTotal = Dinoflagellate total, DiaTotal = Diatom total.
- <0.3 = no correlation;
- >0.3-0.5 = weak correlation, number in underline
- >0.5-0.6 = medium correlation, numbers in italic;
- >0.6-0.7 = strong correlation, number in bold
- >0.7-0.8 = very strong correlation, number in both bold and italic

#### IV. DISCUSSION

##### 1. Environmental factors

The sampling location was strongly affected by different temporal patterns in exchange between two water masses - seawater and brackish estuarine water - most notably during major tidal fluctuations (spring tides). This diurnal water exchange contributed one level of variation in the salinity and nutrient regimes, the precise nature of which was not examined during the present study.

Over the longer seasonal temporal regime of interest, the nutrients ortho-phosphate-phosphorus, nitrate-nitrogen and silicate-silica showed significant variability in concentration, with highest concentrations during the rainy season (low salinity, cool water temperature). The concentrations of ortho - phosphate - phosphorus and silicate-silica at Cua

Be estuary were much higher than those previously reported at Chut Cape, less under the estuarine influence in Nha Trang bay (Fig. 1, Tab. 5, Duong-Trong & Nguyen-Hong 1999). For example, the average concentration of silicate-silica during the present study ( $1024.3 \pm 937.0 \mu\text{g.l}^{-1}$ ) was 3 to 5-fold higher than that at Chut Cape. However, nitrate - nitrogen concentrations were similar between the two locations.

Aquaculture and agriculture along the Cua Be river bank may be the cause for the increase in nutrients at the study site, especially in the rainy season with seasonal cessation of shrimp farming and harvesting of agricultural crops. It is likely that Cua Be estuary receives ortho-phosphate-phosphorus from aqua-culture ponds, and/or rice farms and silicate-silica from natural land via water run-off. However, the similarity in nitrate -

nitrogen concentrations between the two sites suggests either less local influence on this nutrient in the

estuary as compared with Chut Cape, or more widespread dispersal.

**Table 5:** Comparison of the average nutrient concentration at station of Chut Cape and at sampling site of Cua Be estuary, both in Nha Trang bay

NO <sub>3</sub> -N (µg.l <sup>-1</sup> )	PO <sub>4</sub> -P (µg.l <sup>-1</sup> )	SiO <sub>3</sub> -S (µg.l <sup>-1</sup> )	Data of year...	Location	References
174.7 ± 65.9 (n =54)	19.6 ± 15.2 (n = 54)	1024.3 ± 937.0 (n = 54)	Mar. 2000 – Feb. 2001	Cua Be estuary, Nha Trang bay	This study
186.3 107.0	5.6 6.8	181.5 334.0	1997 1998	Chut Cape, out of Cua Be estuary, Nha Trang bay (see map)	Duong-Trong & Nguyen-Hong, 1999

It is well known that nutrient enrichment (e.g. nitrate-nitrogen and ortho-phosphate-phosphorus) of waters from agricultural soils may stimulate blooms of diatoms and dinoflagellates (e.g. Granelli & Moreira 1990). However, in this study, overall phytoplankton biomass exhibited peaks in both the rainy and dry seasons, the latter bearing little apparent relation to nutrient concentrations per se. For most dinoflagellates better growth rates were achieved in the dry season, when concentrations of all nutrients were below their maxima (e.g. nitrate-nitrogen was < 200 µg.l<sup>-1</sup>, phosphate-phosphorous was < 30 µg.l<sup>-1</sup> and silica-

silicate was < 2,000 µg.l<sup>-1</sup>). However, some dinoflagellates (e.g. species of *Peridinium* and *Proto-peridinium*) did reach highest density (785 x10<sup>3</sup> cells.l<sup>-1</sup>) in the rainy season (27 October 2000), immediately after a peak in nitrate-nitrogen and silicate-silica concentrations. Centric diatoms such as *Coscinodiscus* spp. and *Skeletonema costatum* were also dominant during this period, and it seems plausible that this October peak in biomass may have been fostered by the nutrients, as all three nutrients exhibited rapid precipitous decline in concentration in October 2000, immediately preceding high phytoplankton biomass (Tab. 6).

**Table. 6:** Summary of the seasonal variability of biological factors during the present study.

Tabulated values were relative values. Numbers in italic and bracket expressed carbon concentrations (unit = µg.l<sup>-1</sup>)

Season	Gonyaulax (Cells.l <sup>-1</sup> )	Pridiniales (Cells.l <sup>-1</sup> )	Dinoflag. Total (Cells.l <sup>-1</sup> )	Centric diatoms (Cells.l <sup>-1</sup> )	Pennate diatoms (Cells.l <sup>-1</sup> )	Diatom total (Cells.l <sup>-1</sup> )	Phyto. Total (x10 <sup>3</sup> cells.l <sup>-1</sup> )	Chl-a (µg.l <sup>-1</sup> )
Dry	33,000	44,000	236,000 (73)	55,000	1,500	56,500 (13)	292.5 (86)	1.3
Rainy	9,000	59,000	167,000 (56)	98,000	14,000	112,000 (26)	279.0 (82)	1.2
Oct-Nov. (HT- LS)	3,000	102,000	264,000 (91)	169,000	24,000	193,000 (38)	457.0 (129)	1.5
Dec-Feb. (LT- LS)	10,000	3,000	48,000 (18)	17,000	2,000	19,000 (29)	67.0 (47)	1.2
Year Average	27,000	48,000	218,000 (69)	66,000	5,000	71,000 (16)	289.0 (85)	1.3

\* HT-LS = High temperature-Low salinity; LT-LS = Low temperature-Low salinity

## 2. Species composition and cell concentration.

In general, the structure of species composition in the Cua Be river estuary was characterized by a mixture of species from marine, freshwater and brackish water origins with the dominance of *Gonyaulax*, species of Order Peridiniales and species of centric diatoms. In the estuary many of the dominant and common species were present throughout the year, although other species showed strong seasonality (e.g. present only in the dry season). The year-round occurrence of the former species indicates their high tolerances for rapid and episodic shifts in salinity and nutrients (Figs 3, 4 and Appendix Tab. 1).

The year-round occurrence of these species notwithstanding, there was strong seasonal variations in overall species diversity (richness) and composition (Figs. 5, 6), correlated with seasonal trends in temperature and salinity.

These seasonal trends in species diversity and composition were similar to those previously reported in the estuaries and lagoon of Thua Thien-Hue, where dinoflagellates were dominant in the rainy season and diatoms were dominant in the dry season. There, species of *Gonyaulax* and *Protoperdinium* were also characteristic of coastal waters (Nguyen-Ngoc & Doan-Nhu 1997). Conversely, many common Vietnamese dinoflagellate species were notable by their absence in the Cua Be river estuary (e.g. *Ceratium contortum*, *C. falcatum*, *C. gibberum*, *C. horridum*, *C. longirostrum*, *C. karstenii*, *C. lineatum*, *Phalacroma argus*, *P. minutum*, *P. jibbonense*, *P. circumsutum*, *Pyrocystis*

spp.). All are known to occur in Central Vietnam, including Nha Trang bay. Species of *Alexandrium* (e.g. *A. affine*, *A. leei*, *A. tamiyavanichii*, and *A. tamarensis*), were also absent, even though they commonly occur in Nha Trang bay (Nguyen-Ngoc et al. 2002, in press). The reason(s) for the apparent absence from the estuary of this large and common suite of species are not certain, but suggest they may exhibit lower tolerances to fluctuating environmental conditions related to the mixing of estuarine and marine waters (e.g. high turbidity and fluctuations in salinity).

In this study, the total species richness of dinoflagellates and diatoms was less than that previously reported in Nha Trang bay (Hoang-Quoc 1962 & 1963), where 154 species of diatoms and 92 species of dinoflagellates have been described. However, there were no sampling stations located in the Cua Be river estuary in the former study, and the vast majority of the previously listed species were characteristic of tropical coastal waters, not estuaries. The previous work was also limited to two months of sampling (Jan. – Feb. 1961) and thus did not give a general view of species composition or seasonal shifts in community structure as documented in the present study.

There was little difference in total cell concentration between the dry and the rainy seasons and different phytoplankton groups were observed in the two seasons. By contrast, previous studies of phytoplankton in the coastal waters of central Vietnam have shown that average cell concentrations in estuaries, lagoons, and upwelling areas in the rainy season to be 2 times higher than in the dry season (Nguyen-

Ngoc & Doan Nhu 1997).

### 3. Correlations between environmental and biological factors

Most correlations between environmental and biological factors were not clear (Tab. 4). For example, there was no overall relationship between silicate-silica-silica concentration and diatom biomass (also see Gould et al. 1986). Similarly no correlation (or very weak correlation) could be found between phytoplankton abundance (diatoms and dinoflagellates) and nutrients in Tai Tam bay, Hong Kong (Chiu et al. 1994).

In Cua Be river estuary, the highest cell concentration of dinoflagellates occurred in the dry season (high salinity: 32 –35 psu, and temperature: 26-28°C). This is in contrast to earlier studies (e.g. Gould et al. 1986), which found that the highest numbers of dinoflagellates and other algae were associated with lower salinity. Abboud-Abi Saab (1992) also showed that dinoflagellates were more sensitive to fluctuations in salinity than diatoms and significant correlations were limited to certain parameters. For diatoms in Cua Be river estuary, highest cell concentration occurred in low salinity and high silicate-silica concentration, although overall there were no clear correlations among these parameters (Tab. 4). These results also contrast with those of Gould et al. (1986), who found highest diatom concentrations during periods of higher temperature, with low values of dissolved silica.

In conclusion, the phytoplankton community of Cua Be river estuary is comprised of a suite of highly tolerant species that occur year-round and another suite of species that show strong seasonality, associated primarily

with changes in salinity and temperature. The seasonal influences of nutrients were not clear-cut, and it may be that none are particularly limiting, given the extensive aquaculture and agriculture development.

### ACKNOWLEDGEMENTS

This work was a part of PhD programme of Mr. Nguyen Ngoc Lam funded by Danida through an international cooperative program between the ION (Vietnam) and IOC Science & Communication Centre in Harmful Algae, Botanical Institute, Copenhagen University (Denmark). We would like to express our grateful acknowledgement to Dr. Lyndon Devantier from Australian Institute of Marine Science and IUCN Hon Mun MPA Project, for correcting the English language. Mr. Pham Van Thom from the Dept of Hydro-geochemistry and Mr. Tong Phuoc Hoang Son from Dept. of G.I.S. (ION) are thanked by nutrient analysis and suggestion of biological statistical analysis, respectively. Special thanks are also given to Dr. Per Andersen from BioConsult, Denmark for offering the PRIMER Programme and Dr. Vu Do Quynh from Can Tho University (Vietnam) for comments on the statistical analysis. Two reviewers are also thanked.

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**Appendix table 1:** A list of phytoplankton species in Cua Be river estuary recorded in present study, March 2000 – February 2001

TAXA	J	F	M	A	M	J	J	A	S	O	N	D
<b>NOSTOCOPHYCEAE</b>												
1 Oscillatoria sp.										+		
<b>DINOPHYCEAE</b>												
1 Amphisolenia bidentata				+								
2 Ceratium bohmi							+			+		
3 Ceratium furca	+		+	+	+	+	+					+
4 Ceratium fusus	+	+		+		+		+				
5 Ceratium hirundiella											+	+
6 Ceratium trichoceros	+			+			+			+		
7 Ceratium tripos			+	+			+					
8 Dinophysis acuminata		+				+	+					
9 Dinophysis caudata			+		+	+	+					



